IN THE UNITED STATES PATENT AND TRADEMARK OFFICE
BOARD OF PATENT APPEALS AND INTERFERENCES

Atty. Docket No: 018733/0875

In re patent application of
LEUNG, Shui-on *et al.*

Serial No.: 09/185,607

Group Art Unit: 1642

Filed: November 4, 1998

Examiner: L. Helms

For: LANDSCAPED ANTIBODIES AND ANTIBODY FRAGMENTS FOR CLINICAL USE

BRIEF ON APPEAL

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APPELLENTS' BRIEF UNDER 37 CFR §1.192

Commissioner for Patents
P.O. Box 1450
Alexandria, VA 22313-1450

Sir:

This brief is in furtherance of the Notice of appeal filed in this case on April 15, 2003. The fees required under 37 CFR §1.17(f) are herewith included in the attached check. Please charge any fee deficiency or credit any overpayment to Deposit Account 19-0741.

This brief is transmitted in triplicate in conformance with 37 CFR §1.192(a).

I. REAL PARTY IN INTEREST

The real party in interest in this case is Immunomedics, Inc., as evidenced by an assignment filed on January 26, 1999, and recorded at Reel 9727, Frame 0947.

II. RELATED APPEALS AND INTERFERENCES

There are no related appeals or interferences known to appellant, the appellant's legal representative, or assignee which will directly affect or be directly affected by or have a bearing on the Board's decision in the pending appeal.

III. STATUS OF CLAIMS

Pending claims:	1, 2, 4-19, 21-38, 40, 41, 43, 44, 46, 47, 49 and 53-55
Withdrawn claims:	15, 28, 30-37
Canceled claims:	3, 20, 39, 42, 45, 48, 50-52
Claims objected, but allowable:	2, 40 and 43.
Rejected claims:	1, 2, 4-14, 16-19, 27, 29, and 38-55
Appealed claims:	1, 2, 4-14, 16-19, 27, 29, and 38-55

IV. STATUS OF AMENDMENTS

Following a final rejection, an amendment was filed. Subsequently, a request for continued examination was filed which directed that the entry of this amendment into the record. Accordingly, all amendments and arguments are a part of the record for appeal.

V. BACKGROUND AND SUMMARY OF THE INVENTION

Antibody immunoconjugates are widely used in modern medicine. Chemical methods allowing effective conjugation of a variety of diagnostic and therapeutic compounds, including drugs and chelates, to monoclonal antibodies (mabs) are well documented. However, most of these methods rely on random attachments to certain amino acid residues, such as tyrosine, lysine, aspartic acid and glutamic acid. BR-96-DOX 16771 and LL2-*pseudomonas* exotoxin immunoconjugates, which have demonstrated significant anti-tumor activity in tumor-bearing mice, are examples of antibody immunoconjugates constructed through conjugations at these residues. However, because these conjugates are made under extreme chemical conditions (non-physiological pHs, temperature, solvents, *etc.*) and because the conjugation is not site-specific,

the resulting immunoconjugates may exhibit reduced and heterogenous antigen binding properties.[1]

Rodwell *et al.*, *Proc. Nat'l Acad. Sci.*, 83: 2632 (1986), reported site-specific covalent modification of monoclonal antibodies (mAbs) using the Asn-linked carbohydrate (CHO) in the CH2 domain (Asn297) as a convenient chemical handle for radionuclide conjugation. ^{131}I-conjugates formed by this method exhibited homogenous binding properties with improved *in vivo* targeting efficiency in mice. By using a soluble amino-dextran as an intermediate carrier, therapeutic drugs, such as methotrexate (MTX), flourouridine, or doxorubicin (DOX), have been conjugated at the CH2-appended carbohydrate moiety. See, for example, U.S. Patent No. 4,699,784. However, because the Asn297-associated CHO is positioned at the internal space formed between the two adjacent CH2 domains, steric hindrance is expected to impede the efficiency of conjugation at this site. Moreover, antibody fragments, such as $F(ab')_2$, Fab' and Fab, which often are preferred for clinical use, lack the Fc portion and the associated carbohydrate moiety. Accordingly, these species can not be conjugated by this method.[2]

Hansen *et al.*, U.S. Patent No. 5,443,953, and Leung *et al.*, U.S. Provisional Patent Application 60/013,709, the entire contents of which are incorporated herein by reference, describe the introduction of multiple glycosylation sites on the Vκ and CH1 (HCN1 and HCN5 sites) domains of antibodies. Attachment of chelates at all of these sites does not affect the immunoreactivity of the resultant antibody, Leung *et al.*, *J. Immunol.* 154: 5919 (1995), making these carbohydrates ideal site-specific conjugation sites for drugs or chelates. However, in order to conjugate at these carbohydrates, the hexose rings must be chemically oxidized to generate reactive aldehyde groups. Aldehyde groups thus formed can be covalently bonded to the amino groups of chelates or drugs through Schiff bases. Since only the C-C bonds with hydroxyl groups attached to each carbon can be periodate-oxidized to form two aldehyde groups, the

[1] Specification at page 1, lines 20-32.

[2] Specification at page 1, line 33 to page 2, .line 13.

maximum number of these reactive sites is dictated by the structure and linkages of the oligosaccharide.[3]

For example, the compositions and sequences of CH1-appended carbohydrates from two antibodies, hLL2HCN1 and hLL2HCN5, have been determined by fluorophore-assisted carbohydrate electrophoresis (FACE) 16411. Qu *et al.*, *Glycobiol.* 7(6): 803-09 (1997). The structural profile of hLL2HCN1-carbohydrates revealed that about 2-4 hexose rings in an oligosaccharide chain are available for periodate oxidation. Therefore, a maximum of 8-16 aldehyde groups on average can be generated from the carbohydrate side chains of each hLL2HCN1 $F(ab')_2$ fragment. With the average size of hLL2HCN5-carbohydrate being 3-4 monosaccharide residues larger than that of HCN1, a higher number of maximum achievable aldehyde groups for hLL2HCN5 is expected. Under mild chemical conditions, only 1.6 and 3 molecules of DTPA were conjugated to the $F(ab')_2$ of hLL2HCN1 and hLL2HCN5 sites, respectively, probably due to inefficient oxidation of hexose rings under these conditions. Although harsher conditions can be used to generate more aldehyde groups, they may alter the three-dimensional structure of the antibodies and the immunoreactivities of the antibodies may suffer.[4]

Brumeanu *et al.*, *J.Immuno. Meth.* 183: 185-97 (1995), reported coupling peptides to the carbohydrate moieties of antibodies with an enzymatic procedure, in which C-6 aldehydes were generated by oxidizing the terminal galactose (Gal) residues of desialylated immunoglobulins (Igs) with galactose oxidase (GAO). Attachment of peptides is then achieved with concurrent stabilization of the Schiff bases upon mild reduction. The conjugation occurs under physiological conditions, and is specific and efficient with the average number of peptides coupled per Ig being in agreement with the estimated number of galactose equivalents. However, this method requires numerous time consuming steps and cannot be adapted for *in vivo* conjugation in the context of pretargeting.[5]

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[3] Specification at page 2, lines 14-27.

[4] Specification at page 2, line 28 to page 3, line 11.

[5] Specification at page 3, lines 12-21.

There is a need, therefore, for antibodies and antibody fragments that can be conjugated at specific sites to form immunoconjugates useful in clinical applications, such as the diagnosis and treatment of cancer and infectious diseases. There also is a need for a method of making antibody and antibody fragment conjugates wherein the conjugation occurs at specific sites and does not interfere with the specific binding of the antibody or antibody fragment.[6] The present invention meets these needs by providing a glycosylated antibody or antigen-binding antibody fragment having a reactive ketone group on the glycosylated site, wherein said glycosylated site is in the Vκ or CH1 domain, and wherein the reactive ketone group is not introduced by oxidation.[7] Also provided is an immunoconjugate comprising a glycosylated antibody or antigen-binding antibody fragment conjugated to an agent through the glycosylated site, wherein said glycosylated site is in the Vκ or CH1 domain, and wherein the agent is conjugated to a reactive ketone group on the glycosylated site that is not introduced by oxidation.[8]

Methods of making the glycosylated antibodies, antibody fragments and immunoconjugates also are covered. Thus, the application includes a claim directed to a method of making a glycosylated antibody having a reactive ketone group on the glycosylated site, comprising expressing SP2/0 cells that are transfected with a vector encoding an antibody having one or more N-glycosylation sites in the CH1 or Vκ domain in a culture medium comprising a ketone derivative of a saccharide or biosynthetic saccharide precursor, so that they produce a glycosylated antibody having a reactive ketone group on the glycosylated site.[9] The invention also provides a method of making a glycosylated antigen-binding antibody fragment having a reactive ketone group on the glycosylated site comprising expressing SP2/0 cells that are transfected with a vector encoding an antibody having one or more N-glycosylation sites in the CH1 or Vκ domain in a culture medium comprising a ketone derivative of a saccharide or biosynthetic saccharide precursor so that they produce a glycosylated antibody having a reactive ketone group on the glycosylated site, and fragmenting the resulting glycosylated antibody to

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[6] Specification at page 3, lines 22-27.

[7] See claim 19.

[8] See claim 22.

[9] See claim 1.

produce a glycosylated antigen-binding antibody fragment having a reactive ketone group on the glycosylated site.[10] A method of making an immunoconjugate comprising a glycosylated antibody conjugated to an agent through its glycosylated site, comprises reacting a glycosylated antibody produced according to claim 1 or fragment produced according to claim 6 with an agent comprising a ketone-reactive group selected from the group consisting of hydrazides, hydrazines, hydroxylamines, and thiosemicarbazides, thereby conjugating said glycosylated antibody to said agent through the reactive ketone group on its glycosylated site, wherein the reactive ketone group is not introduced by oxidation.[11]

Immunooconjugates according to the invention can be used in a method of targeting an active agent to an *in vivo* target site comprising administering an immunoconjugate comprising a glycosylated antibody or antigen-binding antibody fragment conjugated to an active agent through a reactive ketone group on a glycosylated HCN1, HCN5 or Vκ-N glycosylation site and not as a conjugate to an oxidized sugar.[12]

VI. ISSUES

There are three issues on appeal in the present case:

1. Whether a recitation of "a ketone derivative of a saccharide or biosynthetic saccharide precursor" in claims 1, 4-14, 16-18, 38, 41, 44 and 47 encompasses such "a myriad of compounds" that a skilled artisan would not be able to practice the invention across its full scope. More particularly, the issue is whether compounds other than N-levulinoyl mannosamine and N-levulinoyl fucose are enabled.

2. Whether claims 19, 21-27, 29 and 53-55 would have been obvious based on Shih *et al.* (U.S. 5,057,313) taken in view of Leung *et al.* (*Int. J. Cancer* 60:534-538 (1995)) and Qu *et al.* (*Glycobiology* 7:803-809 (1997)).

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[10] See claim 6.

[11] See claims 8 and16.

[12] See claim 30.

3. Whether the specification reasonably conveyed to one skilled in the relevant art that the inventors had possession of the invention claimed in claims 8-14, 16-18, 19, 21-27, 29, 44, 46, 47, 49, and 53-55 at the time the application was filed. This rejection relates to the recitation "wherein the reactive ketone group is not introduced by oxidation."

VII. GROUPS OF CLAIMS

For purposes of this appeal, the claims do not stand or fall together. The obviousness rejection will argued separately according to the following groups:

Group I Claims 19, 21-27, 29 and 53-55

Group II Claim 27

The first presented arguments under Section IX.B.1. of the brief, apply to all claims rejected under Section 103(a). Then separate arguments are presented under Section IX.B.2. for a claim that recites that the agent is selected from a group of particular ligand-containing peptides (claim 27).

VIII. SUMMARY OF THE ARGUMENT

Contrary to the examiner's assertions, the phrase "a ketone derivative of a saccharide or biosynthetic saccharide precursor" defines "a discrete and identifiable class of compounds," and ***not*** "a myriad of compounds" as alleged by the examiner. The scope of the phrase must be considered in the context of the claimed invention, which is directed to glycosylated antibodies and antibody fragments. There are a very limited number of saccharides that are used in glycosylation. Therefore, claims 1, 4 14, 16 18, 38, 41, 44, and 47 have a well-defined scope and are fully enabled.

Claims 19, 21 27, 29 and 53-55, all of which recite that "the reactive ketone group is not introduced by oxidation,." would not have been obvious based on Shih *et al.* (U.S. 5,057,313) taken in view of Leung *et al.* (*Int. J. Cancer* 60:534-538 (1995)) and Qu *et al.* (*Glycobiology* 7:803-809 (1997)). The cited documents all disclose antibodies in which ketone and/or aldehyde

groups are introduced by oxidation. Furthermore, the specification clearly provides a logical basis why a glycosylated antibody produced according to the present invention will differ from one that is produced by oxidation as in the cited documents..

Finally, the specification reasonably conveys to one skilled in the relevant art that, at the time the application was filed, the inventors had possession of glycosylated antibodies and antibody fragments having "a reactive ketone group that is not introduced by oxidation," as claimed in claims 8 14, 16 18, 19, 21 27, 29, 44, 46, 47, 49, and 53 55. The examiner admits on the record that the specification indeed teaches a method that does not require oxidation. He appears to discount this knowledge, however, because it was gleaned from the background section of the specification. As one of skill in this art, the examiner recognized that the present method does not require oxidation, and was developed to overcome the deficiencies of prior art methods that used oxidation to introduce reactive groups. Thus support for a recitation of "a reactive ketone group that is not introduced by oxidation" is implicit in appellants' disclosure.

IX. ARGUMENT

A. "A ketone derivative of a saccharide or biosynthetic saccharide precursor" as recited in claims 1, 4-14, 16-18, 38, 41, 44, and 47 is fully enabled.

The examiner initially objected to the recitation "ketone derivative of a saccharide or saccharide precursor" under both the first and second paragraphs of Section 112. The rejection under the second paragraph of Section 112 was withdrawn, and currently only a rejection under the first paragraph remains.

More particularly, the examiner asserts that the phrase "a ketone derivative of a saccharide or biosynthetic saccharide precursor" encompasses "a myriad of compounds and the specification only teaches two such compounds of N-levulinoyl mannosamine and N-levulinoyl fucose." This hearkens back to the earlier Section 112 rejection, which urged that "there is no way for a person of skill in the art to ascribe a discrete and identifiable class of compounds to said phrase."

The present invention relates to glycosylated antibodies, and this context provides the skilled artisan with guidance on the scope of the terms questioned by the examiner. There are a very limited number of saccharides that are used in glycosylation. Appended to the response dated December 21, 2001, was an excerpt from Stryer, BIOCHEMISTRY. Figs. 14-18 of that excerpt show the formulae of saccharides commonly found in oligosaccharide units of glycoproteins; these include β-L-fucose, β-D-galactose, β-D-N-acetylgalactosamine, β-D-N-acetylglucosamine, β-D-mannose, and sialic acid. Thus the scope of the term "saccharide" in the context of antibody glycosylation is both well-defined and fully enabled.

The scope of the term "biosynthetic saccharide precursor" is similarly enabled. Taken in the context of glycosylation it is abundantly clear to the skilled artisan that this term refers to molecules that can be converted by biosynthetic pathways in the cell to one of the limited number of saccharides that are used in glycosylation. There are a limited number of saccharide precursors. The specification mentions mannosamine, which can be converted by the cell's biosynthetic machinery to sialic acid, one of the common saccharides used in glycosylation:

> a ketone derivative of a saccharide (such as N-levulinoyl fucose) or saccharide precursor (such as N-levulinoyl mannosamine (ManLev)), resulting in an antibody comprising reactive ketone groups at the N-glycosylation sites. In the case of ManLev, biosynthetic pathways convert the ManLev to levulinoyl sialic acid, which is incorporated into the antibody at the glycosylation site. In the case of N-levulinoyl fucose, the N-levulinoyl fucose itself is incorporated into the antibody at the glycosylation site.

Mannosamine is one of a limited group of well-known saccharide precursors. For example, Yarema *et al.*, of record, describe "[delivery of] a uniquely reactive ketone group to endogenous cell surface sialic acid residues by treating cells with ***the ketone-bearing metabolic precursor N-levulinoylmannosamine (ManLev).***"

Thus, a biosynthetic saccharide precursor is a precursor of one of the saccharides used in glycosylation of glycoproteins, which is converted by the cell's biosynthetic pathways into a saccharide that is incorporated into a glycoprotein. Therefore, the phrase "saccharide or biosynthetic saccharide precursor" *when considered within the context of glycosylated*

antibodies as presently claimed, defines "a discrete and identifiable class of compounds," and ***not*** "a myriad of compounds" as alleged by the examiner.

In the Official Action dated January 15, 2003, the examiner urges that "the claim recites 'a ketone derivative of a saccharide' or 'biosynthetic saccharide precursor' and not that the ketone derivative is a biosynthetic saccharide precursor." This is so, and the claim clearly recites that the ketone derivative is a derivative ***of*** a saccharide or biosynthetic saccharide precursor. A skilled artisan clearly would be apprised that a "ketone derivative" refers to compounds in which a ketone functionality is introduced on a saccharide or biosynthetic saccharide precursor, for example, as an N-acyl group such as levulinoyl, and would be able to introduce such a ketone functionality using nothing more than the level of ordinary skill in the organic synthesis art.

In summary, the examiner's allegation that "a myriad of compounds" are encompassed within the scope of "a ketone derivative of a saccharide or biosynthetic precursor" is ill-founded in the context of glycosylated antibodies as presently recited. Accordingly, the scope of appellant's claims is fully enabled and reversal of the rejection for lack of enablement under the first paragraph of Section 112 is requested.

B. Claims 19, 21-27, 29 and 53-55 would not have been obvious based on Shih *et al.* (U.S. 5,057,313) taken in view of Leung *et al.* (*Int. J. Cancer* 60:534-538 (1995)) and Qu *et al.* (*Glycobiology* 7:803-809 (1997)).

1. All of the references cited by the examiner disclose the use of harsh oxidation conditions to derivatize a glycosylated antibody, and none disclose or suggest a glycosylated antibody or antigen-binding antibody fragment having a reactive ketone group on the glycosylated site that is not introduced by oxidation.

Claims 19, 21-27, 29 and 53-55 are rejected under Section 103(a) based on Shih *et al.* U.S. 5,057,313) in view of Leung *et al.*, *Int. J. Cancer*, 60:534-538 (1995) ("Leung I") and Qu *et al.*, *Glycobiology*, 7:803-809 (1997). The examiner urges that Shih *et al.* discloses oxidizing a carbohydrate of an antibody to produce ketones and conjugating drugs and toxins to the oxidized antibody. Shih *et al.* teaches oxidizing a carbohydrate of an antibody to produce ketones and conjugating drugs and toxins to the oxidized antibody. The examiner admits that Shih *et al.* does not teach antibodies with glycosylation sites at the HCN1, HCN5 or Vκ-N site, but alleges that these deficiencies are made up for by the teaching of Leung I and Qu *et al.*

The claims on appeal recite that "the reactive ketone group is not introduced by oxidation." It is clear that the present application discloses an alternative to introducing reactive groups by oxidizing a sugar. For example, page 2 of the specification discusses Leung *et al.*, *J. Immunol.* 154: 5919 (1995) ("Leung II"), which is exemplary of methods that oxidize a carbohydrate, as follows:

> in order to conjugate at these carbohydrates, the ribose rings must be chemically oxidized to generate reactive aldehyde groups. Aldehyde groups thus formed can be covalently bonded to the amino groups of chelates or drugs through Schiff bases. Since only the C-C bonds with hydroxyl groups attached to each carbon can be periodate-oxidized to form two aldehyde groups, the maximum number of these reactive sites is dictated by the structure and linkages of the oligosaccharide.

The present invention provides glycosylated antibodies that do not require this oxidation, as an alternative to methods like that disclosed in the Leung II and Shih. The present method clearly goes directly from introduction of a ketone derivative onto an antibody to reacting the "resulting antibody" with a ketone reactive group, *i.e.*, the reactive ketone group is introduced as a conjugate to a non-oxidized sugar.

The portion of Shih *et al.* that is cited by the examiner actually is a reference to a published application by McKearn (EP 88,695), which discloses "a method for preparing antibody conjugates which involves oxidizing the carbohydrate portion of the antibody and linking compounds with free amine groups to the resultant carbonyls (aldehyde and/or ketone groups) by Schiff base formation. The harsh oxidation used to open the ring and thereby generate the carbonyl groups, especially where complete oxidation of all carbohydrate residues is desired, and the harsh reducing environment used to stabilize the Schiff base conjugate, both may impair the biological activity of the molecule. By contrast, the antibodies according to the present invention already have a reactive ketone group as a side chain on the carbohydrate used in the glycosylation of the antibody, which is produced by the transfected host cell's biosynthetic machinery. Shih (McKearn) does not disclose a glycosylated antibody or antigen-binding antibody fragment having a reactive ketone group on the glycosylated site that is not introduced

by harsh oxidation. In particular, Shih (McKearn) does not disclose such a glycosylated antibody prepared by the method of claim 1.

The examiner urges that "the claims are directed to a product and there is no indication in the specification or the prior art that the structure of the reactive ketone would be different in the oxidized method versus the biosynthetic method." If the ketone used to derivatize the saccharide or saccharide precursor used for glycosylation is not an oxidized sugar, it is clear that the product is structurally different from the prior art. Furthermore, most oxidized sugars produce aldehydes and not ketones – another clear structural difference from the prior art. Page 3 of the specification explains that the structural profile of hLL2HCN1-carbohydrates reveals that about 2-4 hexose rings in an oligosaccharide chain are available for periodate oxidation, which means that a maximum of 8-16 aldehyde groups on average can be generated from the carbohydrate side chains of each hLL2HCN1 $F(ab')_2$ fragment. With the average size of hLL2HCN5-carbohydrate being 3-4 monosaccharide residues larger than that of HCN1, a higher number of maximum achievable aldehyde groups for hLL2HCN5 is expected. When the oxidizing conditions are mild, only 1.6 and 3 molecules of DTPA can be conjugated to the $F(ab')_2$ of hLL2HCN1 and hLL2HCN5 sites, respectively, due to inefficient oxidation of hexose rings under these conditions. When harsher oxidizing conditions are used to generate more aldehyde groups, the three-dimensional structure of the antibodies is altered and the immunoreactivities of the antibodies may suffer. This would also hold for ketone groups generated by oxidation of glycosylated antibodies. In contrast, the number of glycosylation sites with reactive ketone structures is not limited to those available by oxidation of the oligosaccharide chains. Moreover, the biosynthetic route does not alter the three-dimensional structure of the antibodies. Even if the ketone may not differ in structure, the specification clearly provides a logical basis for why the ***glycosylated antibody*** on which it is contained will be different. Thus, it is reasonable to conclude that the present antibodies are different than those of the prior art.

The addition of Leung I and/or Qu *et al.* to Shih (McKearn) would not have suggested the invention as presently claimed. Leung I discloses a glycosylation site in the Vκ domain and that this site can be used for conjugations. Leung I references other articles which disclose the conjugation technique. Like Shih (McKearn), these entail chemical oxidation of the rings to

generate reactive aldehyde groups, which then can be covalently bonded to the amino groups of chelates or drugs through Schiff bases. Since only the C-C bonds with hydroxyl groups attached to each carbon can be periodate-oxidized to form two aldehyde groups, the maximum number of these reactive sites is dictated by the structure and linkages of the oligosaccharide, hence Leung's disclosure that an average of 2 to 6 chelators such as DTPA could be conjugated.

Qu *et al.* teaches the compositions and sequences of CH1-appended carbohydrates from two antibodies, hLL2HCN1 and hLL2HCN5, as determined by fluorophore-assisted carbohydrate electrophoresis (FACE). The structural profile of hLL2HCN1-carbohydrates revealed that about 2-4 hexose rings in an oligosaccharide chain are available for periodate oxidation. Therefore, a maximum of 8-16 aldehyde groups on average can be generated from the carbohydrate side chains of each hLL2HCN1 F(ab')2 fragment. With the average size of hLL2HCN5-carbohydrate being 3-4 monosaccharide residues larger than that of HCN1, a higher number of maximum achievable aldehyde groups for hLL2HCN5 is expected.

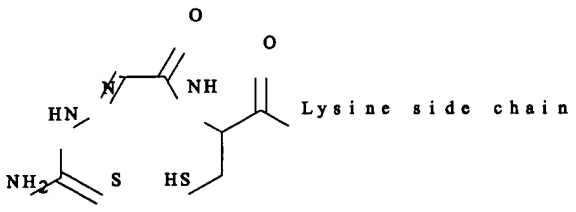
Qu *et al.* does not overcome Shih's failure to teach conjugation methods that use introduced reactive ketone groups on the side chains of the glycosylation carbohydrates, as opposed to chemical oxidation of the carbohydrate ring and subsequent covalent bonding of the thus-generated aldehyde groups to the amino groups of chelates or drugs through Schiff bases. Since only the C-C bonds with hydroxyl groups attached to each carbon can be periodate-oxidized to form two aldehyde groups, the maximum number of these reactive sites is dictated by the structure and linkages of the oligosaccharide. As discussed above, chemical oxidation to generate carbonyl groups has significant adverse consequences. When harsh conditions are used to generate the maximum number of such groups, the three-dimensional structure of the antibodies is altered and the immunoreactivities of the antibodies may suffer. And under milder chemical conditions, only 1.6 and 3 molecules of DTPA are conjugated to the F(ab')2 of hLL2HCN1 and hLL2HCN5 sites, respectively, probably due to inefficient oxidation of hexose rings under these conditions.

All of the references cited by the examiner disclose the use of harsh oxidation conditions to convert a glycosylated antibody to a molecule with available carbonyl functions. The antibodies according to the present invention, on the other hand, have a reactive ketone group on

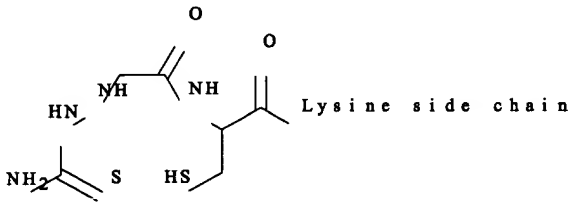
a side chain, and are produced by the transfected host cell's biosynthetic machinery. None of the cited references disclose or suggest a glycosylated antibody or antigen-binding antibody fragment having a reactive ketone group on the glycosylated site that is not introduced by oxidation, and more particularly a glycosylated antibody prepared by the method of claim 1. In accordance with the present invention, these antibodies are made recombinantly by a transfected host cell. The host cell's biosynthetic machinery converts the antibodies so that they have a reactive ketone group. For these reasons, the Board is asked to reverse the rejections under Section 103(a) based on Shih *et al.* in view of Leung I and Qu *et al.*

2. Claim 27 discloses immunoconjugates with particular ligand-containing peptides which are not disclosed in the cited references.

Claim 27 recites an immunoconjugate in which the therapeutic agent is a ligand-containing peptide selected from the group consisting of DTPA-bearing peptides, DOTA-bearing peptides, $AcK_dD_dK_d(TscGC)D_dK_d$-$NH(CH_2)_4CH(NH_2)CONH$-NH_2 and $AcK_dD_dK_d(TsdGC)D_dK_d$-$NH(CH_2)_4H(NH_2)CONH$-NH_2, where K_d and D_d represent the D-amino acids D-lysine and D-aspartic acid, respectively, and where TscGC is the ligand:



and TsdGC is the ligand:



The examiner has identified no teaching in the references of ligands as recited in claim 27, and thus no *prima facie* case of obviousness has been stated with respect to this claim.

C. The specification reasonably conveys to one skilled in the relevant art that, at the time the application was filed, the inventors had possession of glycosylated antibodies and antibody fragments having "a reactive ketone group that is not introduced by oxidation," as claimed in claims 8-14, 16-18, 19, 21-27, 29, 44, 46, 47, 49, and 53-55.

In the latest Official Action the examiner raises a new ground of rejection, alleging that the specification does not reasonably convey to one skilled in the relevant art that, at the time the application was filed, the inventors had possession of glycosylated antibodies and antibody fragments having "a reactive ketone group that is not introduced by oxidation." He admits that "while the specification teaches a method that does not require oxidation, the cited work by Leung II is just prior art and background and the specification does not show support for excluding oxidation of the sugar by chemical methods."

It is well established in the case law that an "appellant's specification need not describe the claimed invention *in ipsis verbis* to comply with the written description requirement . . . The test is whether the originally filed specification disclosure reasonably conveys to a person having ordinary skill that applicant had possession of the subject matter later claimed . . . By the very nature of the inquiry under this statutory provision, each case turns on its own specific facts." *Nelson v. Bowler*, 1 USPQ2d 2076, 2078-2079 (Bd. Pat. App. & Int'f (1986). Similarly, "*ipsis verbis* disclosure is not necessary to satisfy the written description requirement of section 112. Instead, the disclosure need only reasonably convey to persons skilled in the art that the inventor had possession of the subject matter in question." *Fujikawa v. Wattanasin,* 93 F.3d 1559, 1570, 39 USPQ2d 1895, 1904 (Fed. Cir. 1996), and "If a person of ordinary skill in the art would have understood the inventor to have been in possession of the claimed invention at the time of filing, even if every nuance of the claims is not explicitly described in the specification, then the adequate written description requirement is met." *In re Alton,* 76 F.3d 1168, 1175, 37 USPQ2d 1578, 1584 (Fed. Cir. 1996).

The purpose of the written description requirement is to ensure that the applicant has conveyed to those of skill in the art that he or she was in possession of the claimed invention at

the time of filing, and the fundamental factual inquiry regarding the adequacy of disclosure is whether an application conveys with reasonable clarity to those skilled in the art that, as of the filing date sought, applicant was in possession of the claimed invention. *See Vas-Cath, Inc. v. Mahurkar*, 935 F.2d 1555, 1563 (Fed. Cir. 1991). To provide descriptive support, it is not necessary that the application describe the claim limitations exactly. *See e.g. In re Lukach*, 442 F.2d 967, 969 (CCPA 1971) ([T]he invention claimed does not have to be described *in ipsis verbis* in order to satisfy the description requirement of § 112.) Rather, the application need only be sufficiently clear that persons of skill in the art would recognize that the Applicant had possession of the claimed invention. *See In re Wertheim*, 541 F.2d at 263. The description need not be explicit, but may be implicitly or inherently supported in the originally filed disclosure. *Id.* at 1107. Thus, the written description requirement is satisfied when each claim limitation is supported explicitly, implicitly or inherently in the originally filed disclosure. *See* Guidelines for Examination of Patent Applications Under the 35 U.S.C. 112 ¶1, "Written Description" Requirement, 66 Fed. Reg. 1099 (2001). A review of whether the specification complies with the written description requirement is conducted from the standpoint of one of skill in the art at the time the application was filed. *See e.g. Wang Labs. V. Toshiba Corp.*, 993 F.2d 858, 865 (Fed. Cir. 1993).

The examiner admits, for the record, that the specification teaches a method that does not require oxidation. He appears to discount his understanding that this is the case, however, because it was gleaned from the background section of the specification, which describes deficiencies with methods such as those disclosed by Leung *et al.* and other documents that use oxidation to introduce reactive groups the cited work by Leung II. As one of skill in this art, the examiner recognized that the present method does not require oxidation, and was developed in order to overcome the deficiencies of prior art methods that used oxidation to introduce reactive groups. A skilled artisan has recognized that appellants were in possession of glycosylated antibodies and antibody fragments that possess "a reactive ketone group that is not introduced by oxidation," and support for this recitation therefore is implicit in appellants' disclosure. Reversal of the rejection under the first paragraph of Section 112 for lack of written description is requested.

X. CONCLUSION

Based on the foregoing, all claims are believed to be in condition for allowance, and reversal of all grounds for rejection in this case is solicited.

Respectfully submitted,

Dec. 15, 2003
Date

for Stephen B. Maebius
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APPENDIX: PENDING CLAIMS

1. (Twice Amended) A method of making a glycosylated antibody having a reactive ketone group on the glycosylated site, comprising:

expressing SP2/0 cells that are transfected with a vector encoding an antibody having one or more N-glycosylation sites in the CH1 or Vκ domain in a culture medium comprising a ketone derivative of a saccharide or biosynthetic saccharide precursor, so that they produce a glycosylated antibody having a reactive ketone group on the glycosylated site.

2. (Objected to but allowable) The method of claim 1, wherein the ketone derivative of the saccharide or saccharide precursor is selected from the group consisting of N-levulinoyl mannosamine and N-levulinoyl fucose.

3. Canceled

4. The method of claim 1, wherein the antibody has more than one glycosylation site.

5. The method of claim 1, wherein the antibody is a single-chain antibody.

6. (Twice Amended) A method of making a glycosylated antigen-binding antibody fragment having a reactive ketone group on the glycosylated site comprising:

expressing SP2/0 cells that are transfected with a vector encoding an antibody having one or more N-glycosylation sites in the CH1 or Vκ domain in a culture medium comprising a ketone derivative of a saccharide or biosynthetic saccharide precursor so that they produce a glycosylated antibody having a reactive ketone group on the glycosylated site, and

fragmenting the resulting glycosylated antibody to produce a glycosylated antigen-binding antibody fragment having a reactive ketone group on the glycosylated site.

7. The method of claim 6, wherein the fragment is an $F(ab')_2$ fragment.

8. (Twice Amended) A method of making an immunoconjugate comprising a glycosylated antibody conjugated to an agent through its glycosylated site, comprising:

reacting a glycosylated antibody produced according to claim 1 with an agent comprising a ketone-reactive group selected from the group consisting of hydrazides, hydrazines, hydroxylamines, and thiosemicarbazides, thereby conjugating said glycosylated antibody to said agent through the reactive ketone group on its glycosylated site, wherein the reactive ketone group is not introduced by oxidation.

9. The method of claim 8, wherein the antibody is purified before reaction with the agent.

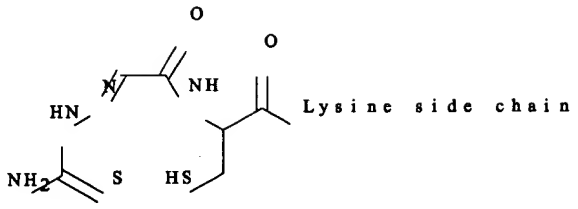
10. The method of claim 8, wherein the agent is added directly to the culture medium.

11. The method of claim 8, wherein the antibody is immobilized on a protein A column prior to reaction with the agent, and eluted from the protein A column after reaction with the agent.

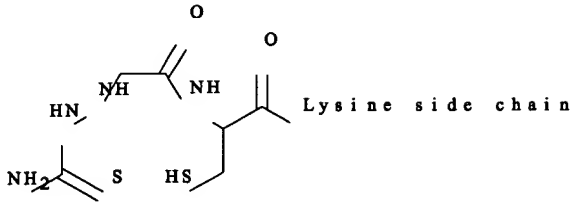
12. The method of claim 8, wherein the agent is selected from the group consisting of diagnostic agents, therapeutic agents, chelating agents and linking agents.

13. The method of claim 12, wherein the agent is selected from the group consisting of peptides, oligosaccharides, biotinamidocaproyl hydrazides, diagnostic markers, drugs, toxins, imaging radioisotopes, and therapeutic radioisotopes.

14. The method of claim 8, wherein the agent is a ligand-containing peptide selected from the group consisting of diethylene triamine pentaacetic acid–bearing (DTPA-bearing) peptides, 1,4,7,10-tetraazacyclododecane-N,N',N"N'"-tetraacetic acid-bearing (DOTA-bearing) peptides, $AcK_dD_dK_d(TscGC)D_dK_d$-$NH(CH_2)_4CH(NH_2)CONH$-NH_2, $AcK_dD_dK_d(TsdGC)D_dK_d$-$NH(CH_2)_4H(NH_2)CONH$-NH_2, and H_2N-NH-CH_2-CO-D_d-$K_d(TscGC)$-D_d-K_d-NH_2, where K_d and D_d represent the D-amino acids D-lysine and D-aspartic acid, respectively, and where TscGC is the ligand:



and TsdGC is the ligand:



15. (Withdrawn) The method of claim 14, wherein the agent is H_2N-NH-CH_2-CO-D_d-K_d(TscGC)-D_d-K_d-NH_2.

16. (Twice Amended) A method of making an immunoconjugate comprising a glycosylated antigen-binding antibody fragment conjugated to an agent through the glycosylated site, comprising:

reacting a glycosylated antibody fragment produced according to claim 6 with an agent comprising a ketone-reactive group selected from the group consisting of hydrazides, hydrazines, hydroxylamines, and thiosemicarbazides, thereby conjugating said glycosylated antibody fragment to said agent through the reactive ketone group on its glycosylated site, wherein the reactive ketone group is not introduced by oxidation.

17. The method of claim 16, wherein the fragment is an $F(ab')_2$ fragment.

18. The method of claim 16, wherein the agent is selected from the group consisting of diagnostic agents, therapeutic agents, chelating agents and linking agents.

19. (Twice Amended) A glycosylated antibody or antigen-binding antibody fragment having a reactive ketone group on the glycosylated site, wherein said glycosylated site is in the Vκ or CH1 domain, and wherein the reactive ketone group is not introduced by oxidation.

20. Canceled.

21. The glycosylated antibody or antigen-binding antibody fragment of claim 19, wherein the antibody or antibody fragment has more than one glycosylation site, each of which has a reactive ketone group.

22. (Twice Amended) An immunoconjugate comprising a glycosylated antibody or antigen-binding antibody fragment conjugated to an agent through the glycosylated site, wherein said glycosylated site is in the Vκ or CH1 domain, and wherein the agent is conjugated to a reactive ketone group on the glycosylated site that is not introduced by oxidation.

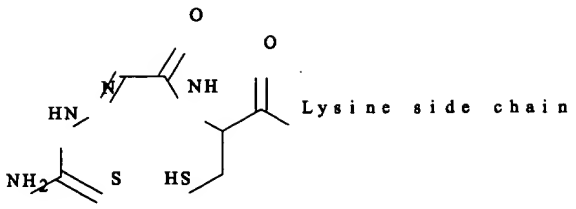
23. The immunoconjugate of claim 22, wherein the glycosylated site is selected from the group consisting of HCN1, HCN5 and Vκ-N.

24. The immunoconjugate of claim 22, wherein the antibody has more than one glycosylated site, each of which is conjugated to an agent.

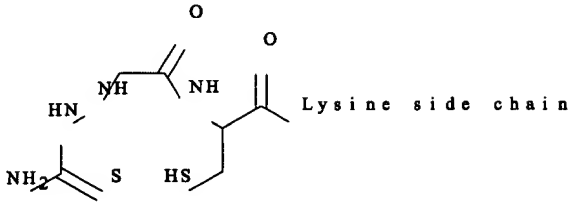
25. The immunoconjugate of claim 22, wherein the agent is selected from the group consisting of diagnostic agents, therapeutic agents, chelating agents and linking agents.

26. The immunoconjugate of claim 25, wherein the agent is selected from the group consisting of peptides, oligosaccharides, biotinamidocaproyl hydrazides, diagnostic markers, drugs, toxins, imaging radioisotopes, and therapeutic radioisotopes.

27. The immunoconjugate of claim 25, wherein the agent is a ligand-containing peptide selected from the group consisting of DTPA-bearing peptides, DOTA-bearing peptides, $AcK_dD_dK_d(TscGC)D_dK_d$-$NH(CH_2)_4CH(NH_2)CONH$-NH_2 and $AcK_dD_dK_d(TsdGC)D_dK_d$-$NH(CH_2)_4H(NH_2)CONH$-NH_2, where K_d and D_d represent the D-amino acids D-lysine and D-aspartic acid, respectively, and where TscGC is the ligand:



and TsdGC is the ligand:



28. (Withdrawn) The immunoconjugate of claim 27, wherein the agent is H_2N-NH-CH_2-CO-D_d-K_d-(TscGC)-D_d-K_d-NH_2.

29. The immunoconjugate of claim 22, wherein the agent is a chelating agent chelated to a diagnostic or therapeutic radioisotope.

30. (Withdrawn) A method of targeting an active agent to an *in vivo* target site comprising administering an immunoconjugate comprising a glycosylated antibody or antigen-binding antibody fragment conjugated to an active agent through a reactive ketone group on a glycosylated HCN1, HCN5 or Vκ-N glycosylation site and not as a conjugate to an oxidized sugar.

31. (Withdrawn) The method of claim 30, wherein the active agent is selected from the group consisting of diagnostic and therapeutic agents.

32. (Withdrawn) The method of claim 30, wherein the antibody or antibody fragment has multiple glycosylated sites, each of which is conjugated to an active agent.

33. (Withdrawn) A method of targeting an active agent to an *in vivo* target site comprising:

administering a glycosylated antibody or antigen-binding antibody fragment having a reactive ketone group on a HCN1, HCN5 or Vκ-N glycosylation site, and allowing the antibody or antibody fragment to localize at the target site;

optionally, administering a clearing agent to clear non-localized antibody or antibody fragment from circulation; and

administering an active agent comprising a ketone-reactive group selected from the group consisting of hydrazides, hydrazines, hydroxylamines, and thiosemicarbazides, whereby the active agent localizes at the target site via conjugation with the pre-targeted antibody or antibody fragment.

34. (Withdrawn) The method of claim 33, wherein the active agent is selected from the group consisting of diagnostic and therapeutic agents.

35. (Withdrawn) The method of claim 33, wherein the clearing agent is administered.

36. (Withdrawn) The method of claim 35, wherein the clearing agent is an anti-idiotypic clearing agent.

37. (Withdrawn) The method of claim 33, wherein the antibody or antibody fragment has more than one glycosylated site, and wherein more than one active agent moiety is conjugated to the pretargeted antibody or antibody fragment.

38. (Amended) A method of making a glycosylated antibody having a reactive ketone group on the glycosylated site, comprising:

expressing SP2/0 cells that are transfected with a vector encoding an antibody having a HCN1, HCN5 or Vκ N-glycosylation site in a culture medium comprising a ketone derivative of a saccharide or biosynthetic saccharide precursor, so that they produce an N-glycosylated antibody having a reactive ketone group on the glycosylated site.

39. Canceled.

40. (Objected to, but allowable) A method according to claim 38, wherein the ketone derivative of the saccharide or biosynthetic saccharide precursor is selected from the group consisting of N-levulinoyl mannosamine and N-levulinoyl fucose.

41. (Amended) A method making a glycosylated antigen-binding antibody fragment having a reactive ketone group on the glycosylated site, comprising:

expressing SP2/0 cells that are transfected with a vector encoding an antibody having a HCN1, HCN5 or Vκ N-glycosylation site in a culture medium comprising a ketone derivative of a saccharide or biosynthetic saccharide precursor, so that they produce a glycosylated antibody having a reactive ketone group on the glycosylated site, and

fragmenting the resulting glycosylated antibody into a glycosylated antigen-binding antibody fragment having a reactive ketone group on the glycosylated site.

42. Canceled.

43. (Objected to, but allowable) A method according to claim 41, wherein the ketone derivative of the saccharide or biosynthetic saccharide precursor is selected from the group consisting of N-levulinoyl mannosamine and N-levulinoyl fucose.

44. (Amended) A method of making an immunoconjugate comprising a glycosylated antibody conjugated to an agent through its glycosylated site, comprising:

reacting a glycosylated antibody according to claim 38 with an agent comprising a ketone-reactive group selected from the group consisting of hydrazides, hydrazines, hydroxylamines, and thiosemicarbazides, thereby conjugating said glycosylated antibody to said agent through the reactive ketone group on its glycosylated site, wherein the reactive ketone group is not introduced by oxidation.

45. Canceled.

46. (Amended) A method according to claim 44, wherein the ketone derivative of the saccharide or biosynthetic saccharide precursor is selected from the group consisting of N-levulinoyl mannosamine and N-levulinoyl fucose.

47. (Amended) A method of making an immunoconjugate comprising a glycosylated antigen-binding antibody fragment conjugated to an agent through the glycosylated site, comprising:

reacting a glycosylated antibody fragment according to claim 41 with an agent comprising a ketone-reactive group selected from the group consisting of hydrazides, hydrazines, hydroxylamines, and thiosemicarbazides, thereby conjugating said glycosylated antibody fragment to said agent through the reactive ketone group on its glycosylated site, wherein the reactive ketone group is not introduced by oxidation.

48. Canceled.

49. (Amended) A method according to claim 47, wherein the ketone derivative of the saccharide or biosynthetic saccharide precursor is selected from the group consisting of N-levulinoyl mannosamine and N-levulinoyl fucose.

50. Canceled.

51. Canceled.

52. Canceled.

53. (Amended) A glycosylated antibody or antigen-binding antibody fragment having a reactive ketone group on a glycosylated site, wherein said glycosylated site is selected from the group consisting of HCN1, HCN5 and Vκ-N, and wherein the reactive ketone group is not introduced by oxidation.

54. (Amended) An immunoconjugate comprising a glycosylated antibody or antigen-binding antibody fragment conjugated to an agent through a reactive ketone on a glycosylated

site, wherein said glycosylated site is selected from the group consisting of HCN1, HCN5 and Vκ-N, and wherein the reactive ketone group is not introduced by oxidation.

55. (Amended) A glycosylated antibody having a reactive ketone group on a glycosylated site, prepared by a method as recited in claim 1, wherein the reactive ketone group is not introduced by oxidation.